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Inter- and intra-specific variation in accumulation of cadmium by peanut, soybean, and navybean

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Abstract. Production of summer grain legumes like peanut, soybean, and navybean is expanding into irrigated or high rainfall areas on more acid, lighter textured soils in coastal areas of north-eastern Australia. A history of intensive use of phosphatic fertilisers, combined with soil properties which generally enhance phytoavailability of cadmium (Cd), have produced concerns about the likely quality of grain legumes produced in these areas. This paper reports field and pot experiments which examine the effect of grain legume species and variety on Cd accumulation when grown across a range of soil types. Results clearly show that both peanut and soybean accumulate Cd in seeds at levels greater than the maximum permitted concentration (MPC, 0.05 mg Cd/kg) even on soils with relatively low total or available Cd concentrations (<0.5 mg/kg). The relative risk of MPC exceedance in marketable seeds or kernels was peanut > soybean > navybean, with the differences between peanut and navybean apparently correlated with differences in total plant Cd uptake. Cadmium concentrations in plant tops always exceeded that in seeds or kernel, and the testa in peanut kernel was shown to contain Cd concentrations that were 50 times greater than that in the embryonic axis and cotyledons.

Significant (P < 0.05) variation in Cd content (at least 2-fold) was recorded among peanut varieties, with lesser variation evident among a limited sample of commercial navybean varieties. Comparison of results for 11 peanut varieties grown at each of 2 locations suggested strong genotype×environment interactions determining kernel Cd concentration.

Highly significant (P < 0.01) linear relationships were established between soil Cd in the cultivated layer (0–20 cm; 0.1 M CaCl₂ extraction) and seed Cd content in field-grown soybean. However, despite observations of an apparent relationship between soil Cd (CaCl₂ extraction) and peanut kernel Cd in pot studies, relationships between soil Cd in the cultivated layer and kernel Cd could not be reproduced in field trials. Kernel Cd concentrations from field-grown peanut plants were generally higher than those from pot trials, despite using soil collected from the cultivated layer (0–20 cm) of the field site for the potting medium. The presence of significant levels of Cd to approximately 60 cm in the soil profile and a general decline in pH_w with depth suggest the lack of correlation between soil test Cd in the top 20 cm and kernel Cd in field-grown plants may be at least partly due to Cd uptake from deeper soil layers.

Additional keywords: grain legumes, Arachis hypogaea, Glycine max, Phaseolus vulgaris, genotypic variation.

Introduction

Major changes are occurring in the cropping systems in which summer grain legumes like peanut (Arachis hypogaea L.), soybean (Glycine max), and navybean (Phaseolus vulgaris) are important components. Traditionally, production of these legumes has centred on rainfed cropping in clay soils with near-neutral pH in water (pH_w, $6 \cdot 0-7 \cdot 5$) and a medium to high clay content that ensures good soil moisture storage. However, industry expansion is occurring into irrigated or high rainfall areas on more acid (pH_w $\leq 5 \cdot 5$), lighter textured soils in coastal areas due to both a need to

stabilise annual production and an interest by growers using traditional monocultures (e.g. sugarcane) to introduce crop rotations to overcome the 'yield decline' syndrome (Garside et al. 1995). Such a shift in industry focus, while guaranteeing greater stability of production of high quality produce, presents an increased chance of product contamination from soil to plant transfer of pesticide and heavy metal residues. In particular, many of these coastal soils have potentially high total Cd concentrations due to the inadvertent addition of high rates of Cd as an impurity in phosphatic fertilisers (Rayment 1997), especially in sugarcane cropping systems which have traditionally been heavily fertilised (Bramley et al. 1996). The combination of low pH_w and light texture in many of the soils of this region would suggest that their affinity for Cd^{2+} is likely to be low and the resultant availability of Cd for uptake by plants (phytoavailability) would be high (Hinesly et al. 1982; McLaughlin et al. 1996).

Recent years have seen an increasing public awareness and concern for food quality, particularly in relation to contamination by pesticide residues and heavy metal contaminants like cadmium (Cd) and lead (Pb). The maximum permitted concentrations (MPCs) of Cd in foodstuffs were established by the National Health and Medical Research Council (NHMRC 1987). Grain legumes fall into the 'all other foods' category in the regulations with an MPC of 0.05 mg/kg, but the regulations are currently under review (Australia and New Zealand Food Authority, pers. comm.).

There are still few published data on Cd concentrations in grain legumes in Australia with the exception of those of Petterson and Harris (1995), who reported Cd concentrations for primarily winter-sown legumes (mainly lupin seed grown in Western Australia). Concentrations of Cd ranged from <0.01 to 0.08 mg/kg in that survey, with few samples exceeding the MPC of 0.05 mg/kg. The authors concluded that there were no significant differences between wheat, field peas, and lupin in their accumulation of Cd, and that problems with excessive Cd accumulation were unlikely with lupin or other legumes grown in that State. However, the authors report no details of soil characteristics at the sites sampled (e.g. clay contents, soil Cd levels, pH, Zn fertility, and salinity status), and all of these factors would have had a major impact on the phytoavailability and uptake of Cd (McLaughlin et al. 1996). It is therefore quite possible that problem areas will exist for Cd uptake, with those areas linked to specific soil characteristics and fertiliser histories.

There is even less Australian information on Cd concentrations in the important summer grain legumes like peanut, soybean, and navybean, although overseas data suggest all are capable of accumulating Cd to levels >0.05 mg/kg under commercial production (e.g. Wolnik *et al.* 1983; Gross *et al.* 1987), especially in soils with high levels of contamination (Stefanov *et al.* 1995). Production which exceeds regulatory or guideline Cd levels is likely to restrict access to both domestic and export markets and should therefore be avoided. This paper reports glasshouse and field studies examining the accumulation of Cd by summer grain legumes and the relationship between crop Cd concentrations and soil properties.

Methods

Field sites

Crop species comparisons were undertaken during the 1994-95 and 1995–96 growing seasons at sites located in the Tully, Ingham, Ayr, Mackay, Bundaberg, and Kingaroy districts on a variety of soil types (Table 1). Soil pH_w ranged from near neutral (Ayr) to moderately acidic (Tully, Ingham, and Mackay), and sites represented a wide range of the lighter textural classes in coastal districts, from the soils at Calavos (4% clay) to those at Ayr and Ingham (26-28% clay). The Kingaroy Red Ferrosol provided a contrasting soil with high clay content typical of the inland rainfed production in traditional growing areas. Crops were either sampled from replicate plots in long-term rotation trials which form part of the Sugarcane Yield Decline Joint Venture (Garside 1997), or from replicate plots in experiments established in commercial fields at Kolan and Calavos near Bundaberg and at Goodger near Kingaroy (Bridge and Bell 1994).

Crops were irrigated at all sites except Kingaroy, Tully, and Ingham, using trickle (Bundaberg rotation trial), flood (Ayr, Mackay), and overhead sprinkler (Calavos, Kolan) systems. Peanuts (Arachis hypogaea L. cv. Streeton) were grown at all sites, and soybeans (Glycine max cv. Leichardt) were sown at Tully (TRT), Ingham (IRT), Ayr (ART), and Mackay (MRT), and cv. Manark was grown at Kingaroy. Various navybean (Phaseolus vulgaris) and limabean (Phaseolus lunatus) varieties were grown at the Bundaberg rotation trial (BRT) and at the Calavos site. In addition, samples of cowpea (Vigna unguiculata cv. Meringa) and maize (Zea mays cv. DK 689) grain were obtained from the TRT and BRT trials, respectively.

Peanut variety trials were undertaken at Kolan and Calavos in the 1994–95 and 1995–96 growing seasons, respectively, to examine genotypic variation in cadmium content of kernel, while similar studies with a small number of commercial navybean varieties were undertaken during the 1995–96 season at both Calavos and the BRT site. The Kolan trial consisted of 30 diverse peanut varieties in a rectangular lattice design with 2 replicates, and the Calavos peanut and navybean variety trials consisted of randomised complete block designs with 3 replicates and 12 or 4 entries, respectively. Five navybean and one Adzuki bean varieties were compared in BRT in a randomised complete block design with 3 replicates.

Glasshouse studies

In order to examine more closely variation in Cd uptake between grain legume species and by crops grown on different soil types, without the possible confounding influences of different depths of effective root-zone commonly observed in field studies, 2 glasshouse pot experiments were undertaken. The first compared Cd uptake by navybean (cvv. Kerman

		values represen	means	of replicate	ed plots from	r each	site			
Location	Great Soil Group ^A	Soil $Classification^B$	pH_w	Org. C^C (%)	${ m CEC}^{ m D}$ $({ m cmol/kg})$	Cl^{E}	$\begin{array}{c} \mathrm{DTPA} \\ \mathrm{Zn}^{\mathrm{F}} \end{array}$	$\begin{array}{c} \text{DTPA} \\ \text{Cd}^{\text{F}} \end{array}$	${{ m EDTA} \atop { m Cd}^{ m G}}$	$\begin{array}{c} CaCl_2 \\ Cd^H \end{array}$
								(mg/l	(mg/kg)	
Tully rotation trial (TRT)	Yellow Earth	Yellow Kandosol	$5 \cdot 4$	$1 \cdot 0$	$2 \cdot 7$	30	$0 \cdot 27$	0.008	0.010	0.009
Ingham rotation trial (IRT)	Soloth	Chromosol	5.5	$1\cdot 2$	$3 \cdot 2$	30	$5 \cdot 15$	$0 \cdot 032$	$0 \cdot 042$	$0 \cdot 032$
Ayr rotation trial (ART)	Alluvial	Melanic Tenosol	$7 \cdot 1$	$1 \cdot 2$	$16 \cdot 4$	50	$2 \cdot 76$	0.040	$0 \cdot 072$	0.005
Mackay rotation trial (MRT)	Solodic	Red Chromosol/ Brown Sodosol	5.3	$0 \cdot 9$	$5 \cdot 0$	10	$1 \cdot 53$	0.039	0.047	$0 \cdot 025$
Bundaberg rotation trial (BRT)	Yellow Podsolic	Bleached Acid Mesotropic Brown Dermosol	6.5	0.9	2.9	20	0.57	0.033	0.058	0.025
Bundaberg– Calavos	Earthy Sand	Acidic Paralythic Orthic Tenosol	5.9	$0 \cdot 6$	$2 \cdot 2$	20	$0 \cdot 27$	0.019	$0 \cdot 029$	$0 \cdot 017$
Bundaberg– Kolan	Red Podsolic	Acid Mottled Mesotropic Red Dermosol	6.0	$0 \cdot 6$	$5 \cdot 2$	20	$0 \cdot 18$	$0 \cdot 029$	$0 \cdot 042$	$0 \cdot 027$
Kingaroy	Kraznozem	Red Ferrosol	$5 \cdot 9$	$1 \cdot 3$	$8 \cdot 1$	15	$3 \cdot 3$	n.d.	$0 \cdot 053$	$<\!0\!\cdot\!005$

 Table 1. Soil classification and selected chemical characteristics from the cultivated layers (0-20 cm) of field experimental sites in coastal Queensland

 Values represent means of replicated plots from each site

n.d., not determined.

^A After Stace *et al.* (1968).

 $^{\rm B}$ After Isbell (1993).

 $^{\rm C}$ Walkeley and Black method (Rayment and Higginson 1992).

 $^{\rm D}$ 1 $\cdot 0$ M ammonium acetate at pH 7 $\cdot 0$ (Rayment and Higginson 1992).

 $^{\rm E}$ Extracted by water at 1:5 soil: water ratio (Rayment and Higginson 1992).

^F Lindsay and Norvell (1978).

^G Clayton and Tiller (1979), modified to 4 h extraction period.

 $^{\rm H}$ 0 $\cdot 1$ $\rm M}$ CaCl_2, 2 h extraction, 1 $: 2 \cdot 5$ soil : solution ratio.

and Spearfelt) and peanut (cvv. Streeton and NC7) varieties grown in large pots (12 kg air-dry soil) containing soil from the cultivated (0–20 cm) layer of the Kolan Red Dermosol. Lime was applied at a rate of 450 mg/kg to achieve a target pH_w of 6.5, and basal fertiliser applications of P (37.3 mg P/kg soil) and K (15 mg K/kg soil) were made using low Cd (<5 mg Cd/kg product) di-ammonium phosphate (DAP) and analytical grade K₂SO₄ for both crop species. Peanut pots received additional topdressed applications of analytical grade CaSO₄ at flowering at a rate of $4 \cdot 8$ g/pot (equivalent to 1.6 g/kg for the soil in the top 10 cm of the pot, approx. 3 kg), while navybeans received a basal application of urea equivalent to 15 mg N/kg soil. Samples taken at both the beginning of reproductive growth and maturity provided information on the relative distribution of Cd between vegetative and reproductive plant parts, as well as a comparison of differences between crop species.

Three replicate pots of each peanut variety were oversown and thinned to achieve stands of 4 plants/pot, with 2 plants destructively sampled at early flowering and the remaining plants grown through to maturity. A similar strategy was followed for the navybean varieties, although only 2 replicate pots were sown to each variety. All pots were free-draining, with watering undertaken using deionised water. All harvested material was washed in deionised water prior to being oven-dried. All plant parts were subsequently ground for analysis with the exception of the peanut pods. These were shelled by hand, with kernels subsequently washed in deionised water and re-dried prior to analysis, and shells were discarded to avoid problems of soil contamination.

A second pot study was undertaken to determine the effects of soil type on Cd uptake in peanut, using soils with varying Cd contents and phytoavailabilities. Soils were collected from the cultivated layers (0–20 cm) of a Kolan Red Dermosol, an adjoining field of Brown Dermosol, and a Red Ferrosol from Kingaroy. All pots received a basal application of low-Cd DAP (<5 mg Cd/kg product) at a rate equivalent to $37 \cdot 3$ mg P/kg soil and an application of analytical grade KCl to provide $12 \cdot 5$ mg K/kg. Pots also received an application of analytical grade CaSO₄ at a rate of $1 \cdot 6$ g/kg at flowering to ensure adequate calcium availability for kernel development. There were 3 replicate pots for each soil type, with plants oversown and thinned to achieve a density of 1 plant/pot. Other cultural and analytical techniques were similar to those employed in the first pot study.

Soil and plant analyses

Plant material (tops) was oven dried (60°C) and both tops and seeds were ground using a stainless steel grinder. Samples were totally digested in a concentrated solution of HNO₃/H₂O₂, after which the solution Cd concentration was determined by graphite furnace atomic absorption spectrophotometry (GF– AAS). Certified reference materials were analysed routinely to ensure that these techniques were adequate. Analyses of NBS wheat (Ref. No. 1567A, certified Cd concentration of 0.026 ± 0.002 mg/kg) and NIES rice flour (Ref. No. 10B, certified Cd concentration of 0.32 ± 0.02 mg/kg) standards gave Cd concentrations of 0.028 ± 0.001 and 0.333 ± 0.009 mg/kg, respectively, during the course of the studies.

In the case of analyses of peanut kernel, whole kernels (with testa intact) were analysed except in the instance where the relative Cd contents of testa and seed were determined. All plant Cd concentrations are expressed on a dry weight basis, and those of seeds are on an 'as received' basis, with moisture contents ranging from 2 to 5%.

Soil pH_w, electrical conductivity (EC), and extractable Cl were determined in a water suspension of soil using a 1:5soil: solution ratio (Rayment and Higginson 1992). Chloride in the filtered solution was determined using an automated ferricyanide method (APHA, 1992). Organic C was determined using the Walkeley-Black procedure (Rayment and Higginson 1992). EDTA-extractable Cd was determined by shaking soils for 4 h in 0.05 M EDTA (pH 6.0) using a soil: solution ratio of 1:5 (modified Clayton and Tiller, 1979). DTPA-extractable Cd was determined by shaking soils for 2 h in 0.005 M DTPA using a soil: solution ratio of 1:2 (Lindsay and Norvell 1978). EDTAand DTPA-extractable Cd represent strongly and moderately strongly surface-bound Cd, respectively. CaCl₂-extractable Cd (weakly bound surface Cd) was determined by extracting soils for 2 h in 0.1 M CaCl₂ solution using a soil:solution ratio of $1:2\cdot 5$ (modified Sauerbeck and Styperek 1985). Concentrations of Cd in extracts were determined using flame or GF-AAS with deuterium background correction.

Statistical analyses

Standard analysis of variance techniques were used to compare varietal differences in Cd accumulation in both the peanut and navybean field trials, and the effects of soil type on Cd accumulation by peanut cv. Streeton in the second pot trial. Paired *t*-tests were used to compare Cd concentrations between plant parts and between navybeans and peanuts in the first pot trial.

Least squares linear regressions were used to examine relationships between soil test Cd and seed or kernel Cd, using individual replicate or bulk sample (in the case of Kingaroy) data from the various experimental sites where common crop varieties were sown.

Results

Species and varietal comparisons: field study

Cadmium concentrations in peanut kernel were in excess of the current MPC (0.05 mg/kg) at all sites except Kingaroy, whereas in soybean seed the MPC was exceeded only at the IRT site (Table 2). Cadmium concentrations in peanut kernels were 4–7 times the MPC in some of the Bundaberg crops, whereas navybean

samples were always <0.05 mg/kg. The differences between species were often large. At Calavos there was a 10-fold difference in Cd concentrations between peanut kernel and navybean seed, whereas at TRT and BRT there were similar order of magnitude differences between Cd concentrations in peanut and cowpea and peanut and maize, respectively.

Table 2. Effect of crop species on grain Cd concentration (mg/kg) in a series of field experiments in coastal Queensland Data are presented as means (±s.e.) for each location. There were 3 replicates at each site except for Bundaberg (Kolan), where there were only 2 replicates

Location	Crop species	Variety	Grain Cd^A
Tully rotation trial	Peanut	Streeton	0.082 ± 0.008
(TRT)	Soybean	Leichardt	0.022 ± 0.003
	Cowpea	Meringa	< 0.009
Ingham rotation trial	Peanut	Streeton	0.083 ± 0.006
(IRT)	Soybean	Leichardt	0.061 ± 0.003
Ayr rotation trial	Peanut	Streeton	$0.055 {\pm} 0.013$
(ART)	Soybean	Leichardt	0.019 ± 0.002
Mackay rotation trial	Peanut	Streeton	0.060 ± 0.003
(MRT)	Soybean	Leichardt	0.041 ± 0.002
Bundaberg rotation	Peanut	Streeton	0.101 ± 0.005
trial (BRT)	Navybean	Sirrius	< 0.009
	Maize	DK 689	< 0.009
Bundaberg–Calavos	Peanut	Streeton	$0.348 {\pm} 0.068$
0	Navybean	Sirrius	0.033 ± 0.003
Bundaberg–Kolan	Peanut	Streeton	0.178 ± 0.020
Kingarov ^A	Peanut	Streeton	$0 \cdot 011$
<i>.</i>	Soybean	Manark	$0 \cdot 021$

^A Analysis of bulk samples only.

Concentrations of Cd in soils varied at least 5-fold across the sites, irrespective of the reagent used to extract Cd from the soil (Table 1). There was a good correlation $(r^2 > 0.94, P < 0.001)$ between concentrations of CaCl₂-extractable Cd in the cultivated layer of the profile (0-20 cm) and Cd concentrations in soybean seed (Fig. 1a), with the only apparent departure from the linear relationship being a single plot at the IRT site. In contrast, there were no significant relationships between soybean seed Cd and extractable Cd in soil determined using either EDTA $(r^2 = 0.09, \text{ n.s.})$ or DTPA $(r^2 = 0.19, \text{ n.s.})$ extracts. The soil samples from each rotation trial site represented a bulked sample taken from within each replicate, rather than from the specific plot in which the soybean crop was grown. The soil Cd concentrations in the sample from replicate 1 at Ingham were half those in the other 2 replicates, although this was not reflected in the seed Cd contents. Considerable spatial variation in Zn levels had also been noted during site characterisation at this location (data not shown), particularly within the replicate from which this sample was obtained (S. Berthelsen, pers. comm.), so this data point was not included in regression calculations.



Fig. 1. Relationship between 0.1 M CaCl₂-extractable Cd in the top 20 cm of the soil profile and (a) cv. Leichardt soybean seed Cd from trials at Tully, Ingham, Ayr, and Mackay or (b) peanut kernel Cd across all rotation trial sites and commercial fields. Data points represent individual replicate plot values for both soil and seed or kernel Cd.

There were no apparent relationships between kernel Cd in peanuts and soil Cd determined by $CaCl_2$ extraction (Fig. 1*b*), or by any other method. Of particular interest were the very high kernel Cd levels recorded at the Kolan and Calavos sites, and the low values at Kingaroy. The low Cd contents of both soybean and peanut seeds grown at Kingaroy were consistent with the very low CaCl₂-extractable Cd levels typical of this soil type. However, the high levels at the Bundaberg sites were recorded despite soil Cd levels in the cultivated layer, which were only in the mid-range.

Differences in Cd contents among varieties of both peanut and navybean were also large, especially for peanut (Table 3). For simplicity we present only the 1995–96 data for peanut from Calavos and navybean/limabean from BRT, although the 1994–95 data from the experiments at Kolan provided an even larger relative variation in Cd contents among peanut varieties (0.16-0.50 mg Cd/kg). Both crop species showed at least 2-fold variation in Cd concentrations among varieties, despite the quite low levels recorded for the *Phaseolus* sp. varieties at BRT.

A comparison of the Cd concentrations in kernels of peanut varieties common to both the Kolan and Calavos studies (Fig. 2) suggests that there was considerable genotype×environment interaction for kernel Cd concentration. An example of this was cv. RMP 91, which recorded the lowest Cd concentrations of all 30 varieties at Kolan in 1994–95 ($0.158\pm0.079 \text{ mg Cd/kg}$) but the highest Cd concentrations ($0.667\pm0.029 \text{ mg}$ Cd/kg) at Calavos in 1995–96. On a more positive note, the current commercial cv. Streeton consistently produced the lowest kernel Cd of all the currently available varieties, and the foliage disease resistant parent cv. Tifrust recorded the lowest kernel Cd in both experiments.



Fig. 2. Comparison of kernel Cd content of peanut varieties grown in both the 1994–95 (Kolan) and 1995–96 (Calavos) trials at Bundaberg. Vertical (1994–95) and horizontal (1995–96) bars indicate l.s.d. values (P = 0.05) for each experiment.

$Glasshouse \ studies$

The species differences in Cd uptake expected from field studies were clearly evident when comparing Cd concentrations in plant tops at both flowering and

Table	3.	Variation	\mathbf{in}	\mathbf{mean}	\mathbf{Cd}	concentration	(mg/	/kg)	of	seeds	of	peanut,	navybean,	and
			li	mabeaı	ı va	rieties grown a	t Bu	ndab	erg	g in 19	96-	96		

The peanut variety trial was grown at Calavos, and the *Phaseolus* sp. variety trial was conducted as part of the Bundaberg rotation trial

Variety	Seed or kernel Cd	Variety	Seed or kernel Cd
Peanut		Pean	uut (cont.)
RMP 91	0.667	B57 P4 1	0.289
A140 L31	$0 \cdot 471$	Tifrust	0.284
Florunner	$0 \cdot 460$		
New Mexico	$0 \cdot 446$	Phased	olus vulgaris
Valencia C		(Ne)	avybean)
NC7	$0 \cdot 441$	Kerman	0.019
Southern	$0 \cdot 404$	Rainbird	$0 \cdot 012$
Runner		Spearfelt ^A	$0 \cdot 012$
B57 P5 1	0.351	Actolac ^A	$0 \cdot 007$
55-437	0.334	Sirrius ^A	$0 \cdot 005$
Q24168	0.333	Phased	olus lunatus
Streeton	0.326	(Li	mabean)
A166 L17	0.307	$\operatorname{Bridgeton}^{A}$	0.006
Peanut l.s.d. $(P = 0.05)$	$0 \cdot 0$	86	
Phaseolus sp. l.s.d. $(P = 0.05)$	n.s	3.	

n.s., not significant.

^A Indicates varieties in which some replicate values were less than the report limit (0.009 mg/kg). For statistical purposes, such samples were assigned Cd contents approx. half the report limit (i.e. 0.005 mg/kg).

maturity, with peanut accumulating Cd in significantly greater concentrations (P < 0.05, Table 4). However, the large differences expected between species in seed Cd at maturity were not evident, although peanut kernels still contained Cd at twice the concentration found in navybean seeds. The contrast between Cd concentrations in peanut kernel from these pot studies and from those recorded in the field was marked. Concentrations in kernel from potted plants $(0.059\pm0.011 \text{ mg/kg})$ were much lower than those recorded in the field for both Streeton $(0.18\pm0.02 \text{ mg/kg})$ and NC7 $(0.31\pm0.09$ mg/kg) on the same soil type. Chemical analyses of soil used in these pot studies (data not shown) showed little difference from the data recorded for the field site and shown in Table 1, with the exception of a higher pH_w in the potted soil $(pH_w \ 6.7)$ than in the field $(pH_w 5.9)$ due to an application of lime as part of the basal fertiliser. This had been done to ensure good navybean growth in this soil.

Differences in Cd concentrations between tops and seeds were more pronounced in peanut than navybean, although in both species Cd in tops exceeded Cd in seeds (Table 4). Navybean also showed a trend for lower Cd concentrations in seeds than in the surrounding pod, although this difference was not statistically significant. Whilst similar analyses were not undertaken for peanut in this study due to problems with soil adhering to the peanut pod, analyses of the Cd concentrations in the skin (testa) and remaining bare kernel on samples of cv. Streeton from the BRT site (0.101 mg Cd/kg, Table 2) showed that a significant proportion of the total kernel Cd was contained in the testa $(2.94\pm0.09 \text{ mg Cd/kg})$ compared with the remaining bare kernel $(0.058\pm0.003 \text{ mg Cd/kg})$.

The second pot experiment (Table 5) examined the effects of soil type on the Cd concentration in peanut kernel. Kernel Cd levels differed significantly

Table 4. Accumulation of Cd (mg/kg) and distribution between vegetative and reproductive plant parts in two varieties of both navybean and peanut grown in pots in the glasshouse

Soil was collected from the cultivated layer of the Bundaberg– Kolan Red Dermosol. Values are shown as means (\pm s.e.), with n = 2 (navybeans) or n = 3 (peanut)

Variety	Plant t	ops at:	Pods	Mature kernels or seeds	
·	Flowering	Maturity			
		Peanut			
Streeton	0.349	$0 \cdot 223$	n.d.	0.061	
	$(0 \cdot 026)$	$(0 \cdot 082)$		$(0 \cdot 011)$	
NC7	$0 \cdot 217$	0.180	n.d.	0.057	
	$(0 \cdot 059)$	$(0 \cdot 019)$		$(0 \cdot 014)$	
		Navybea	n		
Kerman	$0 \cdot 072$	$0 \cdot 058$	$0 \cdot 046$	$0 \cdot 032$	
	$(0 \cdot 011)$	$(0 \cdot 027)$	$(0 \cdot 011)$	$(0 \cdot 008)$	
Spearfelt	0.083	$0 \cdot 111$	0.044	0.036	
-	$(0 \cdot 009)$	$(0 \cdot 062)$	$(0 \cdot 014)$	$(0 \cdot 019)$	

n.d., not determined.

	values followed by different letters are significantly different $T = 0.00$								
Soil	$\mathrm{p}\mathrm{H}_{\mathrm{w}}$	${ m EC} m (mS/cm)$	Cl (mg/kg)	$EDTA-Cd^A$	$CaCl_2-Cd^B$	Total Cd^{C}	Kernel Cd		
Kingaroy Red Ferrosol	$6 \cdot 7$	$0 \cdot 07$	12	0.059	$<\!0\!\cdot\!0015$	$0 \cdot 067$	$0 \cdot 007a$		
Bundaberg–Kolan Brown Dermosol	$6 \cdot 1$	$0 \cdot 06$	26	$0 \cdot 023$	$0 \cdot 003$	$0 \cdot 032$	$0 \cdot 094 \mathrm{b}$		
Bundaberg–Kolan Red Dermosol	$5 \cdot 2$	$0 \cdot 30$	222	0.039	$0 \cdot 0055$	$0 \cdot 044$	$0 \cdot 185c$		

 Table 5. Mean kernel Cd concentrations (mg/kg) of peanut cv. Streeton and selected chemical characteristics of three soils used in the second glasshouse pot trial

Values followed by different letters are significantly different P = 0.05

^A Clayton and Tiller (1979).

 $^{\rm B}$ 0.01 m CaCl₂, 4 h extraction, 1:5 soil:solution ratio.

^C Zarcinas *et al.* (1996).

(P < 0.05) among soil types, ranging from 0.007 mg Cd/kg in the Kingaroy Ferrosol to 0.185 mg Cd/kg in the Kolan Red Dermosol. In contrast to results from the field sites (Fig. 1b), differences were strongly correlated with CaCl₂-extractable soil Cd. This result suggested that the poor relationships between soil test Cd and kernel Cd in field studies at a range of different sites (Fig. 1b) may have been due to factors other than the appropriateness of the soil extract to predict available Cd for this crop species.

The differences in kernel Cd concentrations of cv. Streeton grown in a Bundaberg–Kolan Red Dermosol in this study (0.185 mg Cd/kg) compared with that in the species comparison study (0.061 mg Cd/kg: Table 4) could not be determined. However, there were notable differences in both the pH_w and chloride status of the soils used in the respective studies (Tables 1 and 5). These differences were due to basal applications of lime in the trial comparing crop species and the use of muriate of potash (KCl) as the fertiliser K source for the study comparing soil types.

Discussion

Data presented in this paper clearly show that a number of important summer grain legume species can accumulate Cd in seeds at concentrations greater than the MPC, given appropriate combinations of soil properties and only relatively moderate levels of soil Cd compared with other crop species (Rayment 1997). Exceedance of the MPC would severely limit market access for such produce, so a high likelihood of occurrence of Cd contamination provides a serious impediment to industry expansion into most established irrigation areas with acidic, lighter textured soils. Of particular concern are the very high levels that can accumulate in peanut under field conditions (5–10 times the MPC in some cases: Table 3), even at low Cd concentrations in soil (viz. EDTA or DTPA-extractable Cd < 0.075 mg/kg).

The effects of soil type on Cd content of seeds, both in the field (Table 2) and the glasshouse (Table 5), were generally consistent with expected effects of soil properties (pH, organic matter status, and contents of clay and anhydrous iron and aluminium oxides) on Cd phytoavailability (Chaney and Hornick 1978; McLaughlin et al. 1996). The high affinity of iron and a luminium oxides for retention of Cd^{2+} (Tiller et al. 1984; Rayment 1997), in combination with the moderate organic matter status typical of cultivated krasnozems in the tropics and subtropics (Moody 1994), should maintain low activities of Cd^{2+} in the soil solution. This was reflected in the extremely low ratio of CaCl₂-Cd/total Cd and low levels of Cd in seeds and kernels on the Kingaroy Ferrosol (Tables 1 and 5). Conversely, the lower organic matter contents and lighter textures of most of the coastal sites resulted in higher ratios of CaCl₂-Cd/total Cd and higher seed Cd contents. These results suggest that the easiest way to avoid Cd contamination in grain legume crops is to select soils with low Cd phytoavailability.

The strong effects of both species and variety on Cd concentrations in seeds or kernels of grain legumes were also important. Choice of crop species for soils with low to moderate levels of phytoavailable Cd can have a major impact on the marketability of seed produced. with the apparent risks of MPC exceedance in the order peanut > soybean > navybean (Table 2). However, even navybean seeds can exceed the MPC under certain circumstances and there have been a number of recordings of commercial crops with Cd concentrations of 0.05-0.06 mg Cd/kg from acidic, sandy soils in northern Queensland (Bean Growers Australia, pers. comm.). The reasons for species differences in seed Cd concentrations require further examination, but the pot trial results (Table 4) suggest that differences are consistent with differences in plant Cd uptake, rather than a difference in translocation and/or exclusion of Cd from seeds. Indeed, the observations of greater Cd

concentrations in plant tops than in seeds or kernels and the observations of greater Cd concentrations in outer layers of peanut kernel are both consistent with observations in other species (Kubota *et al.* 1992; Oliver *et al.* 1993).

The generally higher seed Cd concentrations for peanut and navybean recorded in the field, particularly the former crop (Table 2 v. Tables 4 and 5), are unexpected and may be due to a number of factors. Generally, concentrations of metals in plants grown in pots under glasshouse conditions are greater than in the same plants grown in identical soils in lysimeters or in field plots (de Vries and Tiller 1978). This has been explained as resulting from differences between pot and field growth conditions relating to root exploration, soil temperature, and moisture effects, all of which impact on metal uptake by plants (Chaney and Hornick 1978). It is therefore unusual to find higher Cd concentrations in plants grown under field than glasshouse conditions, given identical soils. Possible reasons for this difference are: use of poorer quality irrigation water under field conditions mobilising soil Cd (McLaughlin et al. 1994); different rooting patterns in the field with roots exploring zones of higher Cd phytoavailability; or higher pH_w in the potted soils due to addition of limestone. The first hypothesis is unlikely as irrigation water quality in the areas investigated here is excellent (generally <120 mg Cl/L). The second hypothesis is a likely explanation as analysis of soils to depth (100 cm) at the Bundaberg sites have indicated little change with depth in concentrations of EDTA-extractable Cd to 40 cm, after which there is a decline to levels below the limits of detection at depths >60 cm. These Cd profiles are typically associated with a fall in pH_w from near $6\cdot 0$ in the cultivated layer to $4 \cdot 5 - 5 \cdot 0$ at 60 cm (M. J. Bell and M. J. McLaughlin, unpubl. data), which should increase the relative phytoavailability of soil Cd with depth (Chaney and Hornick 1978). The last hypothesis is also likely, at least in the pot trial comparing peanut and navybean (Table 4), as soil pH_w in the glasshouse experiments was increased from $5 \cdot 9$ to $6 \cdot 7$ by addition of $CaCO_3$, to ensure good navybean growth.

The combination of phytoavailable Cd present in the 20–60 cm depth strata and greater root activity by peanut than by *Phaseolus* sp. (in particular) and soybean at low pH (Wade *et al.* 1988) suggest that for peanut there is greater potential for significant Cd uptake from below the cultivated layer than for other grain legumes. This contribution could be exacerbated by inadequate soil moisture availability in the surface soil, as occurs under rainfed conditions or when irrigation applications are insufficient to meet crop demand. This is consistent with the poor correlations between soil test Cd in the cultivated layer and kernel Cd in peanut (Fig. 1b) and the relatively high Cd levels recorded at Kolan and Calavos (Table 2), where irrigation frequencies were often inadequate during the 1994–95 and 1995–96 seasons (Peter Hatfield, pers. comm.). The strong genotype×environment interactions in Cd concentrations in kernels from different peanut varieties (Fig. 2) may also be related to varietal differences in root activity with depth, and further studies are needed in this area.

Effects of fertility management for a given soil type were illustrated by comparing Cd contents of cv. Streeton grown in the 2 pot trials (Tables 5 and 7). Both studies used the cultivated layer of a Red Dermosol from Kolan, but differing pH_w (due to lime applications to remove limitations to navybean growth) and soil chloride contents (due to contrasting K fertiliser sources) combined to produce a 3-fold variation in kernel Cd levels (0.061 mg Cd/kg in Table 5 to 0.185 mg Cd/kg)in Table 7). Such variation is encouraging in that it suggests that field agronomic management strategies (e.g. liming, fertiliser sources, and irrigation water quality) may be developed to minimise Cd contents of produce grown on a given soil type. Effects of pH amendment (reviewed by McLaughlin et al. 1996) and K fertiliser form (Sparrow et al. 1994) on Cd content of produce have been reported in other crop species, although the effectiveness of such measures seems to be generally lower in the field than in pot or solution culture studies (McLaughlin et al. 1993; Tiller et al. 1994).

The development of models to allow prediction of seed Cd content from soil properties determined by soil tests prior to planting would be of considerable benefit to grain legume industries seeking to evaluate potential new production areas. Our data for soybean cv. Leichardt (Fig. 1a) hold particular promise for that crop in that a simple relationship may exist between concentrations of CaCl₂-extractable Cd in the cultivated layer and seed Cd concentration, although the relationship was developed over only 4 sites (albeit on contrasting soil types) and therefore requires further validation. Similar strong correlations between CaCl₂-extractable Cd and seed or plant Cd have been obtained in other species (e.g. lettuce, Andrewes et al. 1996; celery, Rayment 1997), although these were also over a limited range of sites and soil types. Models relating soil Cd concentrations to crop Cd concentrations are often complicated combinations of soil Cd status (often at multiple depths in the profile) and other soil properties (e.g. chloride concentration, Li et al. 1994; McLaughlin et al. 1994), pH (Jackson and Alloway 1992), or concentrations of Zn (McLaughlin et al. 1994) or organic C (He and Singh 1993) in soil.

Cadmium accumulation in summer grain legumes

Finally, our data on the very high Cd concentrations present in the peanut testa (commonly only 2–3% of the total kernel weight) of cv. Streeton kernel from the BRT field site are of major significance for both producers and processors. The vast majority of peanuts consumed in Australia have the testa removed (Peanut Company of Australia, unpubl. data), with this process having a significant impact on Cd content of the edible product. In the case of the BRT sample of cv. Streeton, testa removal resulted in a virtual halving of the total kernel Cd content (from 0.101 to 0.058 mg Cd/kg). The approximate 50-fold increase in Cd concentration in the testa, relative to the embryonic axis and cotyledons it encloses, suggests that this membrane may represent a relatively efficient barrier to Cd penetration. Wagner (1993) discusses a number of possible mechanisms which could facilitate these phenomena. This issue requires further investigation in terms of the extent to which Cd concentrations in the testa can increase relative to the Cd concentration of the overall kernel sample and the potential for exploitable genotypic variation in testa Cd accumulation. More definitive studies of the mechanisms involved in Cd retention in the testa are also needed.

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References

- Andrewes, P., Town, R. M., Hedley, M. J., and Loganathan, P. (1996). Measurement of plant-available cadmium in New Zealand soils. Australian Journal of Soil Research 34, 441–52.
- APHA (1992). 'Standard Methods for the Examination of Water and Waste Water.' 18th Edn. (American Public Health Association: Washington DC.)
- Bramley, R. G. V., Ellis, N., Nable, R. O., and Garside, A. L. (1996). Changes in soil chemical properties under long-term sugar cane monoculture and their possible role in sugar yield decline. *Australian Journal of Soil Research* 34, 967–84.

- Bridge, B. J., and Bell, M. J. (1994). Effect of cropping on the physical fertility of krasnozems. Australian Journal of Soil Research 32, 1253–73.
- Chaney, R. L., and Hornick, S. B. (1978). Accumulation and effects of cadmium on crops. *In* 'Proceedings, First International Cadmium Conference, San Francisco'. pp. 125–40. (Metals Bulletin Ltd: London.)
- Clayton, P. M., and Tiller, K. G. (1979). A chemical method for the determination of heavy metal content of soils in environmental studies. CSIRO Australia Division of Soils, Technical Paper No. 41.
- Garside, A. L. (1997). Soil components of yield decline in sugarcane. In 'Proceedings, International Symposium on Advances in Soil Quality for Land Management: Science, Practice and Policy'. (Eds R. J. MacEwan and M. R. Carter.) Univ. Ballarat, 17–19 April 1996. pp. 93–6. (University of Ballarat: Ballarat.)
- Garside, A. L., Magarey, R. C., and Nable, R. O. (1995). Growth of different plant species in fumigated/sterilised and untreated sugarcane soils with varying cropping histories. *Proceedings Australian Society of Sugar Cane Technologists* 17, 123–7.
- Gross, R., Auslitz, J. Schramel, P., and Payer, H. D. (1987). Other elements in seeds of *Lupinus mutabilis* and of other legumes. *Journal of Environmental Pathology, Toxicology* and Oncology 7, 59–66.
- He, Q. B., and Singh, B. R. (1993). Effect of organic matter on the distribution, extractability and uptake of cadmium in soils. *Journal of Soil Science* 44, 641–50.
- Hinesly, T. D., Redborg, K. E., Ziegler, E. L., and Alexander, J. D. (1982). Effects of soil cation exchange capacity on the uptake of cadmium by corn. *Soil Science Society of America Journal* 46, 490–7.
- Isbell, R. F. (1993). A classification system for Australian soils (third approximation). CSIRO Australian Division of Soils, Technical Report No. 2/1993.
- Jackson, A. P., and Alloway, B. J. (1992). The transfer of cadmium from agricultural soils to the human food chain. *In* 'Biogeochemistry of Trace Metals'. (Ed. D. Adriano.) pp. 109–158. (Ann Arbor Science: Ann Arbour, MI.)
- Kubota, J., Welch, R. M., van Campen, D. R., and Van Campen, D. R. (1992). Partitioning of cadmium, copper, lead and zinc amongst above-ground parts of seed and grain crops grown in selected locations in the USA. *Environmental Geochemistry and Health* 14, 91–100.
- Li, Y-M., Chaney, R. L., and Schneiter, A. A. (1994). Effect of soil chloride level on cadmium concentration in sunflower kernels. *Plant and Soil* 167, 275–80.
- Lindsay, W. L., and Norvell, W. A. (1978). Development of a DTPA soil test for zinc, iron, manganese and copper. Soil Science Society of America Journal 42, 421–8.
- McLaughlin, M. J., Maier, N. A., Williams, C. M. J., Tiller, K. G., and Smart, M. K. (1993). Cadmium accumulation in potato tubers—occurrence and management. *In* 'Proceedings, 7th National Potato Research Workshop'. (Ed. J. Fennell.) pp. 208–13. Ulverstone, May 1993. (Tasmanian Department of Primary Industry: Launceston.)
- McLaughlin, M. J., Tiller, K. G., Beech, T. A., and Smart, M. K. (1994). Increased Soil salinity causes elevated cadmium concentrations in field-grown potato tubers. *Journal* of Environmental Quality 34, 1013–18.
- McLaughlin, M. J., Tiller, K. G., Naidu, R., and Stevens, D. P. (1996). Review: the behaviour and environmental impact of contaminants in fertilisers. *Australian Journal of Soil Research* 34, 1–54.

- Moody, P. W. (1994). Chemical fertility of krasnozems: A review. Australian Journal of Soil Research 32, 1015–41.
- NHMRC (1987). 'NHMRC Food Standards Code.' (Australian Government Publishing Service: Canberra.)
- Oliver, D. P., Gore, P. J., Moss, M. H., and Tiller, K. J. (1993). Cadmium in wheat-grain and milling products from some Australian flour mills. Australian Journal of Agricultural Research 44, 1–11.
- Petterson, D. S., and Harris, D. J. (1995). Cadmium and lead content of lupin seed grown in Western Australia. Australian Journal of Experimental Agriculture 35, 403–7.
- Rayment, G. E. (1997). Sources of cadmium in agricultural products. Invited paper to National Cadmium Workshop, May 1995. (Dept. Primary Industries and Energy/Australia and New Zealand Food Authority: Canberra.)
- Rayment, G. E., and Higginson, F. R. (1992). 'Australian Laboratory Handbook of Soil and Water Chemical Analyses.' (Inkata Press: Sydney.)
- Sauerbeck, D. R., and Styperek, P. (1985). Evaluation of chemical methods for assessing the Cd and Zn availability from different soils and sources. *In* 'Chemical Methods for Assessing Bio-available Metals in Sludges and Soils'. (Eds R. Leschber, R. D. Davis, and P. L'Hermite.) pp. 49–66. (Elsevier Applied Science Publishers,: London.)
- Sparrow, L. A., Salardini, A. A., and Johnstone, J. (1994). Field studies of cadmium in potatoes (*Solanum tuberosum* L.). III. Response of cv. Russet Burbank to sources of banded potassium. *Australian Journal of Agricultural Research* 45, 243–9.
- Stace, H. C. T., Hubble, G. D., Brewer, R., Northcote, K. H., Sleeman, J. R., Mulcahy, M. H., and Hallsworth, E. G. (1968). 'A Handbook of Australian Soils.' (Rellim Technical Publications: Glenside, S. Aust.)
- Stefanov, K., Seizova, K., Yanishlieva, N., Marinova, E., and Popov, S. (1995). Accumulation of lead, zinc and cadmium in plant seeds growing in metalliferous habitats in Bulgaria. *Food Chemistry* 54, 311–13.

- Tiller, K. G., Gerth, J., and Bruemmer, G. (1984). The relative affinities of Cd, Ni and Zn for soil clay fractions and goethite. *Geoderma* 34, 17–35.
- Tiller, K. G., Oliver, D. P., McLaughlin, M. J., Merry, R. H., and Naidu, R. (1994). Managing cadmium contamination of agricultural land. *Advances in Environmental Science* (in press).
- de Vries, M. P. C., and Tiller, K. G. (1978). Sewage sludge as a soils amendment, with special reference to Cd, Cu, Mn, Ni, Pb and Zn—comparison of results from experiments conducted inside and outside a glasshouse. *Environmental Pollution* 16, 231–40.
- Wade, M. K., Gill, D. W., Subagjo, H., Mohammed Sudjadi, and Sanchez, P. A. (1988). Overcoming soil fertility constraints in a transmigration area of Indonesia. Tropsoils Bulletin 88–01. Soil Management Collaborative Research Support Program, Raleigh, NC, USA. (North Carolina State University: Raleigh, NC.)
- Wagner, G. J. (1993). Accumulation of cadmium in crop plants and its consequences to human health. Advances in Agronomy 51, 173–212.
- Wolnik, K. A., Fricke, F. L., Capar, S. G., Braude, G. L., Meyer, M. W., Satzger, R. D., and Bonnin, E. (1983). Elements in major raw agricultural products in the United States. I. Cadmium and lead in lettuce, peanuts, potatoes, soybeans, sweet corn and wheat. *Journal of Agriculture and Food Chemistry* **31**, 1240–4.
- Zarcinas, B. A., McLaughlin, M. J., and Smart, M. K. (1996). The effect of acid digestion technique on the performance of nebulisation systems used in inductively coupled plasma spectrometry. *Communications in Soil Science and Plant Analysis* 27, 1331–54.

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